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Original Research

# Effect of *Aloe vera* gel on antioxidant enzymes in streptozotocin-induced cataractogenesis in male and female Wistar rats

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## Abstract

**Background:** There is increasing evidence that complications related to diabetes are associated with increased oxidative stress. *Aloe vera* (AV) gel has several biological properties, including antioxidant activity.

**Purpose:** This study was undertaken to evaluate the effects of AV gel extract on oxidative stress in streptozotocin (STZ)-induced diabetic rats.

**Methods:** Wistar albino male and female rats were divided into the following four groups: normal control rats (N), AV-treated normal (N + NT), diabetic (DU), and AV-treated diabetic (DU + DT). The AV-treated normal (N + NT) and AV-treated diabetic (DU + DT) groups received oral administration of AV gel extract (300 mg/kg body weight) for 60 days. Diabetes was induced experimentally by an intraperitoneal injection of STZ at a dose of 55 mg/kg body weight.

**Results:** By the end of the experimental period, levels of various biochemical parameters such as superoxide dismutase, catalase, lipid peroxidation (LPO), protein oxidation (POD), glutathione peroxidase (GPx), glutathione reductase, Aldose reductase (AR), were increased, whereas the level of reduced glutathione (GSH) and Sorbitol dehydrogenase (SD), soluble protein, and insoluble protein were decreased under diabetic conditions. Oral administration of AV gel extract at a dose of 300 mg/kg body weight for 60 days resulted in the prevention of the aforementioned abnormalities.

**Conclusion:** In conclusion, our data demonstrate the protective role of AV leaf extract in inhibiting STZ-induced diabetic oxidative stress, and therefore, this plant could be used as an adjuvant agent for the prevention and/or management of diabetes and aggravated antioxidant status. Copyright © 2014, Taiwan Society of Emergency Medicine. Published by Elsevier Taiwan LLC. All rights reserved.

**Keywords:** *Aloe vera*; antioxidants; glutathione; oxidative stress; streptozotocin

## 1. Introduction

Diabetes is currently growing at a rapid rate throughout the world. In addition, diabetes is the 16th leading cause of global mortality,<sup>1</sup> and diabetic cataract is reported to be an underlying cause of blindness in nearly half of the world's blind population.<sup>2</sup> Cataract is characterized by cloudiness or opacification of the crystalline eye lens. Traditionally, the cataract

intervention program is evaluated by the number of cataract operations performed per million population per year (Vision 2020). However, the surgery has its own limitations, including more pronounced postoperative inflammatory response, loss of vitreous humor, posterior capsule, opacification,<sup>3</sup> and in addition it is expensive.<sup>4</sup> Thus, there is a need not only to look at the impact of treating cataracts and relate it just to surgery but also to look at scholastic achievements and development in the management of cataract due to diabetes. Sex differences have also been reported in the severity of experimentally induced diabetic cataract. However, there are some studies in which this case has not been observed, perhaps due to differences in the site of streptozotocin (STZ) administration or

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dosage. In recent years, the role of alternative therapeutic approaches has become very popular<sup>5</sup> and because various plant extracts may have many pharmacological activities (e.g., antidiabetic, antioxidant, and antistress), they can be effectively used to delay or counter diabetic complications such as cataract.

For this study, we selected *Aloe vera* (AV) as this was already mentioned in ayurveda for the treatment of diabetes mellitus and has been reviewed elsewhere.<sup>6</sup> In brief, AV (kalabanda in Telugu and coastal aloe in English) belongs to the Liliaceae family, of which there are approximately 360 species.<sup>7</sup> Over the years, the plant has been known by a number of names such as “the wand of heaven”, “heaven’s blessing”, and the “silent healer”. They are succulent perennials, which secrete a watery juice from the tubular cells that run lengthwise throughout the stout and fleshy leaves. The mucilaginous tissue in the center of the AV leaf is called gel.

The objective of this investigation was to evaluate the effects of AV leaf extract in delaying or preventing diabetic cataract. Therefore, this study was designed to investigate the protective effect of AV leaf gel extract on the levels of lens lipid peroxide (LPO) and enzymatic antioxidants in male and female rats with STZ-induced diabetes.

## 2. Materials and methods

### 2.1. Chemicals

STZ, DL-glyceraldehyde, and 2-thiobarbituric acid were obtained from Sigma-Aldrich (St. Louis, MO, USA). All other chemicals and solvents were of analytical grade and were obtained from Sisco Research Laboratories (P) Ltd. (Mumbai, India).

### 2.2. Preparation of AV gel extract

AV leaves were collected from the Department of Botany, Sri Krishnadevaraya University (Anantapur, Andhra Pradesh). Mature, healthy fresh leaves were washed with water and cut transversely into pieces. The thick epidermis was selectively removed. The solid gel was then homogenized and the resulting mucilaginous, thick, straw-colored homogenate was stored in dry sterilized small containers at 4°C until further use. The gel was freshly prepared every time and administered orally. The dosing schedule was once per day.

### 2.3. Animals

Male and female Albino Wistar rats (130–160 g) used in the present study were procured from Sri Venkateshwara Enterprises (Bangalore, India). The animals were acclimatized for 7 days in our animal house (Registered number: 470/01/a/CPCSEA) before dietary manipulation. All the animals had free access to water. Food intake (daily) and body weight (weekly) were monitored regularly. Animal care and study protocols were in accordance with and approved by the Institutional Animal Ethics Committee.

### 2.4. Experimental design

At the time of dietary manipulation, all the animals were 6 weeks of age, weighing approximately 200 g. The rats were divided into four groups, with 10 animals in each group as follows: Group N, normal rats (N); Group NT, normal rats treated with AV leaf gel (300 mg/kg/day; NT), Group DU, rats with STZ-induced diabetes that received a single intraperitoneal injection of STZ (55 mg/kg) in 0.1 mL of 0.05M citrate buffer at pH 4.5 (DU), and Group DT, diabetic rats treated with AV leaf gel (300 mg/kg/day; DT).

### 2.5. Lens collection and processing

After the experimental period, the animals were fasted overnight and euthanized by cervical decapitation. The lenses were dissected by the posterior approach and stored at –70°C until further analysis. A 10% homogenate mixture was prepared from three to four pooled lenses in 50mM potassium phosphate buffer at pH 6.2. All the biochemical parameters were analyzed in the soluble fraction of the lens homogenate (centrifuged at 16,000g at 4°C) except for lens malondialdehyde (MDA) and reduced glutathione (GSH), which were determined in the total homogenate.

### 2.6. Biochemical estimations

The extent of lipid peroxidation was determined by assaying MDA formation according to the method suggested by Utley et al.<sup>8</sup>

The protein carbonyl content was measured by forming hydrazone derivatives using 2,4-dinitrophenylhydrazine, which were quantified spectrophotometrically at 370 nm according to the method suggested by Levine et al.<sup>9</sup>

The total GSH content was measured by following the method suggested by Ellman.<sup>10</sup> This method is based on the development of a yellow color, which is due to the reaction of 5,5'-dithio-2-nitrobenzoic acid with the compounds containing sulfhydryl groups. The maximum absorbance was observed at 412 nm.

Tyrosine and tryptophan present in the proteins react with Folin–Ciocalteu reagent in the presence of alkaline copper to give a colored complex with maximum absorbance at 750 nm.<sup>11</sup>

The lysate was prepared from the lens of rats from different experimental groups. The enzyme preparation was allowed to react with H<sub>2</sub>O<sub>2</sub> in the presence of GSH for a specified period according to the method recommended by Rotruck et al.,<sup>12</sup> and the remaining GSH was measured by following the method suggested by Ellman.<sup>10</sup>

Glutathione reductase (GR) catalyzes the reduction of oxidized glutathione by nicotinamide adenine dinucleotide phosphate-reduced (NADPH) to GSH. The activity of the enzyme was measured by following the oxidation of NADPH spectrophotometrically at 340 nm according to the method suggested by Pinto and Bartley.<sup>13</sup>

The activity of glutathione-S-transferases (GSTs) was measured by monitoring the increase in the absorbance at 340 nm using 1-chloro-2,4-dinitrobenzene as a substrate according to the method suggested by Habig et al.<sup>14</sup>

Catalase (CAT) catalyzes the breakdown of  $H_2O_2$  to  $H_2O$  and  $O_2$  and the rate of decomposition of  $H_2O_2$  was measured spectrophotometrically at 240 nm following the method suggested by Beers and Sizer.<sup>15</sup>

The activity of superoxide dismutase (SOD) was measured based on the ability of the enzyme to inhibit the autoxidation of pyrogallol. A modified procedure followed by Soon and Tan<sup>16</sup> was adopted in this study.

The activity of sorbitol dehydrogenase (SDH) was measured by following the method of Asada and Galambos.<sup>17</sup> The SDH catalyzes the reduction of fructose to sorbitol in the presence of nicotinamide adenine dinucleotide-reduced as the reducing agent. The activity was measured by monitoring the decrease in absorbance at 340 nm using fructose as the substrate.

Aldose reductase (AR) catalyzes the NADPH-linked reduction of aldose. The activity was measured by monitoring the decrease in absorbance at 340 nm using glyceraldehyde as a substrate according to the method suggested by Hayman and Kinoshita.<sup>18</sup>

Ascorbic acid is oxidized by copper to form dehydroascorbic acid and 2,4-dinitrophenyl hydrazone, which then undergoes rearrangement to form a product with absorption maximum at 520 nm.<sup>19</sup>

## 2.7. Statistical analysis

Duncan's multiple range test<sup>20</sup> was used for testing statistical significance between groups of data. All results are expressed as mean  $\pm$  standard error of the mean of 10 animals in each group. A  $p$  value  $< 0.05$  was taken to be significant.

## 3. Results

### 3.1. Food intake and body weights

There was an increase in the food intake in the diabetic group (Group DU) compared with the control group (Group N). Despite the increased food intake, the body weight of Group DU decreased (185.8 g for male rats and 157.5 g for female rats at 75 days), when compared with the control group (310 g for male rats and 217.5 for female rats at 75 days). The decrease in body weight due to hyperglycemia was rectified by treatment with AV gel extract (data not shown).

### 3.2. Molecular basis for the delay of cataract

To investigate the possible mechanism by which AV gel extract delays the STZ-induced diabetic cataract, various biochemical parameters related to cataractogenesis such as oxidative stress/antioxidant system, the polyol pathway, protein carbonyl content, and protein content were studied. In

addition, blood glucose and insulin levels were also estimated (data not shown).

### 3.3. Oxidative stress markers and antioxidant system

There was a significant elevation in thiobarbituric acid reactive substances (TBARSs) during diabetes (male 61.5% and female 44%) compared with the normal group, which indicated increased LPO levels in the lens due to hyperglycemia (Fig. 2). Protein carbonyl content was significantly higher in the diabetic group (male 125% and female 944%) compared with the control group, suggesting enhanced protein oxidation under hyperglycemic condition (Fig. 3). In addition, we have also observed a decreased level of GSH in Group DU compared with Group N (Fig. 1). Treatment with gel extract prevented the alterations in TBARSs (male 25.3% and female 26.5%) and protein carbonyl content (male 39.2% and female 22.2%), and improved the GSH content beyond the normal values indicating the beneficial effect of AV gel. The activities of GSH-dependent enzymes, namely, GR (male 75.3% and female 33.4%) and glutathione peroxidase (GPx; (male 16.6% and female 48.5%) decreased in Group DU compared with Group N indicating that there is an increased oxidative stress in diabetic cataract lenses. The beneficial effect of administration of AV gel was observed when the levels of GSH-dependent antioxidant enzymes (i.e., GR and GPx) in Group DT rats were restored to normal (male 32% and female 16%, male 15.5% and female 0.71% respectively). No change was observed in the activity of GSTs. The decreased ascorbic acid levels in Group DU (male 13.2% and female 9%) compared with the control group was rectified by the treatment with AV gel (male 0.6% and female 3.2%; Tables 1 and 2). The activities of SOD and CAT were decreased in Group DU (male 31.7% and female 27.5%, male 64.8% and female 35.6% respectively) compared with Group N. Treatment with AV gel prevented the altered activities of these enzymes (male 31.1% and female 3.7%, male 15.7% and female 6.2% respectively; Tables 1 and 2).

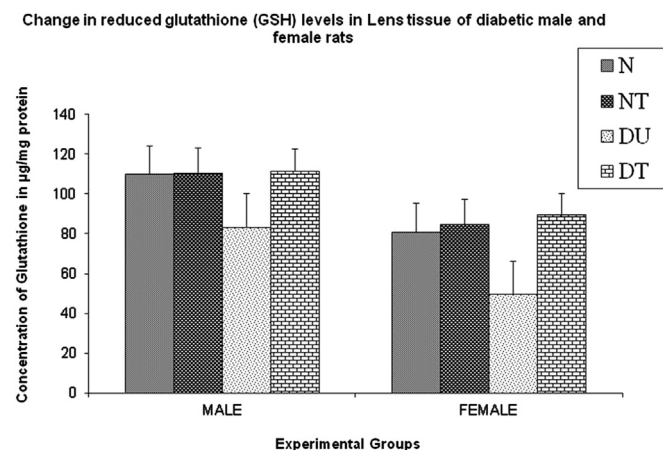


Fig. 1. Change in the levels of reduced glutathione in the lens tissues of diabetic rats after 60 days of treatment with *Aloe vera* gel extract. Data are mean  $\pm$  standard error ( $n = 10$ ). Data are significant at  $p < 0.05$  compared with the diabetic control group.

Change in Lipid peroxidation (LPO) levels in Lens tissue of diabetic male and female rats

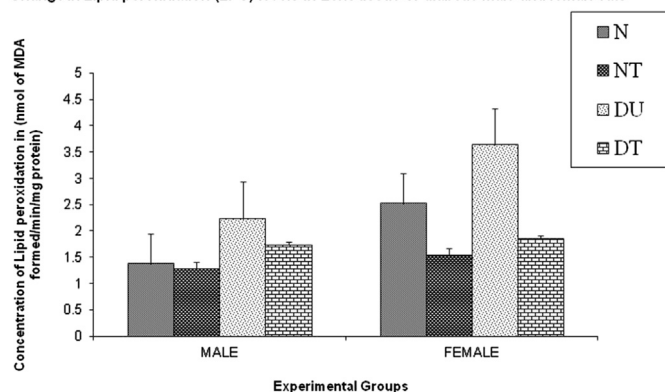


Fig. 2. Change in the levels of lipid peroxidation (MDA) in the lens tissues of diabetic rats after 60 days of treatment with *Aloe vera* gel extract. Data are mean  $\pm$  standard error ( $n = 10$ ). Data are significant at  $p < 0.05$  compared with the diabetic control group.

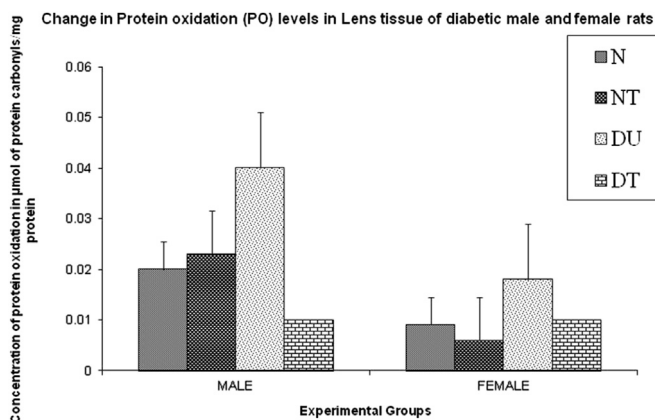


Fig. 3. Change in the levels of protein oxidation in the lens tissues of diabetic rats after 60 days of treatment with *Aloe vera* gel extract. Data are mean  $\pm$  standard error ( $n = 10$ ). Data are significant at  $p < 0.05$  compared with the diabetic control group.

### 3.4. Polyol pathway

The specific activity of AR, a key enzyme in polyol pathway, was significantly higher in Group DU animals (male 85.3% and female 290.9%) than in Group N. The SDH activity decreased in Group DU (male 49.1% and female 36.6%) compared with Group N (Tables 3 and 4). Treatment with AV gel rectified the alterations in polyol pathway enzymes (male 10.7% and female 17.4%, male 8.9% and female 1.2% respectively).

### 3.5. Protein content

Data on the total soluble and insoluble protein content revealed significant decreases in both soluble (male 30% and female 73.9%) and insoluble protein (male 28.1% and female 9.8%) in Group DU compared with Group N (Tables 5 and 6). Feeding the rats with AV gel improved the soluble (male 57.6% and female 10.4%) and insoluble (male 0.6% and female 16.4%) protein levels.

## 4. Discussion

Cataract is an important complication of diabetes mellitus responsible for blindness worldwide. Epidemiological studies have reported a positive association between cataract and diabetes mellitus. It has been estimated that a delay in cataract onset by 10 years could reduce the need for cataract surgery by as much as half.<sup>21</sup> Keeping in view that cataract formation is a slow process and for the purpose of its prevention, drugs will have to be continued for a long period, drugs of natural origin may be an agent of choice owing to their comparative safety, especially if used as foods. Plants such as *Ocimum sanctum* and *Camellia sinensis* were reported to possess antioxidant properties and offer protection against cataract.<sup>22,23</sup> In this study, the AV gel extract delayed the progression of cataract in rats. Although multiple mechanisms may contribute to these effects, the antioxidant effect of AV gel appears to be the predominant mechanism of action.

Oxidative stress plays an important role in the pathogenesis of cataract. The increased TBARSs and protein carbonyls

Table 1

Effect of *Aloe vera* treatment on lens antioxidants in male rats with STZ-induced diabetes. Please define “CDNB” and “DMRT” in tables.

Parameter	N	NT	DU	DT
Glutathione reductase ( $U^a$ /mg protein)	2.43 $\pm$ 0.16	2.59 $\pm$ 0.16	0.60 $\pm$ 0.21*	1.65 $\pm$ 0.25**
Glutathione peroxidase ( $U^b$ /mg protein)	6.19 $\pm$ 0.13	7.15 $\pm$ 1.74	5.1 $\pm$ 0.54*	5.23 $\pm$ 0.78**
Glutathione-S-transferase ( $\mu$ mol of GSH–CDNB conjugate formed/min/mg protein)	0.007 $\pm$ 0.00	0.007 $\pm$ 0.00	0.002 $\pm$ 0.00*	0.005 $\pm$ 0.00**
Superoxide dismutase ( $U^c$ /mg protein)	7.71 $\pm$ 0.39	7.40 $\pm$ 1.86	5.26 $\pm$ 0.93*	5.31 $\pm$ 1.44
Catalase (mmol of $H_2O_2$ decomposed/min/mg protein)	1.33 $\pm$ 0.16	1.29 $\pm$ 0.24	0.46 $\pm$ 0.17*	1.12 $\pm$ 0.40**
Ascorbic acid	1.51 $\pm$ 0.11	1.61 $\pm$ 0.13	1.31 $\pm$ 0.16*	1.50 $\pm$ 0.02**

Values are presented as mean  $\pm$  standard error of the mean for 10 rats in each group.

CDNB = chloro-dinitro benzene; DMRT = Duncan multiple range test; GSH = reduced glutathione; NADPH = nicotinamide adenine dinucleotide phosphate-reduced; STZ = streptozotocin.

\*Significant as compared with Group DU ( $p < 0.05$  DMRT).

\*\*Significant as compared with Group DT ( $p < 0.05$  DMRT).

<sup>a</sup> Micromole of NADPH oxidized/minute.

<sup>b</sup> Microgram of GSH consumed/minute.

<sup>c</sup> Amount of enzyme that gave 50% inhibition of pyrogallol autooxidation/minute.



Table 2  
Effect of *Aloe vera* treatment on lens antioxidants in female rats with STZ-induced diabetes.

Parameter	N	NT	DU	DT
Glutathione reductase ( $U^a$ /mg protein)	2.12 ± 0.32	2.30 ± 0.22	1.41 ± 0.08*	1.78 ± 0.09
Glutathione peroxidase ( $U^b$ /mg protein)	4.18 ± 0.67	4.28 ± 0.21	2.15 ± 0.37*	4.21 ± 0.32**
Glutathione-S-transferase ( $\mu$ mol of GSH–CDNB conjugate formed/min/mg protein)	0.004 ± 0.00	0.004 ± 0.00	0.004 ± 0.00	0.005 ± 0.00
Superoxide dismutase ( $U^c$ /mg protein)	5.54 ± 0.82	4.22 ± 0.73	4.04 ± 0.46	5.33 ± 0.43
Catalase (mmol of $H_2O_2$ decomposed/min/mg protein)	0.73 ± 0.05	0.86 ± 0.12	0.47 ± 0.14	0.78 ± 0.23
Ascorbic acid	1.55 ± 0.08	1.57 ± 0.07	1.41 ± 0.05	1.60 ± 0.11**

GSH = reduced glutathione; NADPH = nicotinamide adenine dinucleotide phosphate-reduced; STZ = streptozotocin.

Values are presented as mean ± standard error of the mean for 10 rats in each group.

\*Significant as compared with Group DU ( $p < 0.05$  DMRT).

\*\*Significant as compared with Group DT ( $p < 0.05$  DMRT).

<sup>a</sup> Micromole of NADPH oxidized/minute.

<sup>b</sup> Microgram of GSH consumed/minute.

<sup>c</sup> Amount of enzyme that gave 50% inhibition of pyrogallol autoxidation/minute.

Table 3  
Effect of *Aloe vera* treatment on lens polyol pathway enzymes in male rats with STZ-induced diabetes cataract.

Parameter	Tissue	Male rats			
		N	NT	DU	DT
Aldose reductase ( $\mu$ mol of NADPH oxidized/min/mg protein)	Cataractous lens	2.32 ± 0.21	2.41 ± 0.16	4.30 ± 0.20*	2.57 ± 0.28**
Sorbitol dehydrogenase ( $\mu$ mol of NADH oxidized/min/mg protein)	Cataractous lens	2.34 ± 0.28	2.40 ± 0.25	1.19 ± 0.17*	2.13 ± 0.33**

NADH = nicotinamide adenine dinucleotide-reduced; NADPH = nicotinamide adenine dinucleotide phosphate-reduced; STZ = streptozotocin.

Values are presented as mean ± standard error of the mean for 10 rats in each group.

\* Significant as compared with Group DU ( $p < 0.05$  DMRT).

\*\* Significant as compared with Group DT ( $p < 0.05$  DMRT).

along with the decreased GSH and altered activities of anti-oxidant enzymes and vitamin C in this study suggest increased oxidative stress in diabetic conditions. No variation was observed in the activity of GR in the N rats of both genders, whereas significantly higher activity of GPx was observed in male N rats than female rats. Previous studies reported that hyperglycemia induces polyol pathway that consumes NADPH, which is necessary for GPx redox cycle.<sup>24</sup> Decreased GPx activity in diabetic rats is probably due to insufficient GSH levels. In diabetes, disturbances in ascorbic acid metabolism might have a great role in the pathogenesis of diabetic complications.<sup>25</sup> In this study, decreased vitamin C levels were observed in diabetic rats, which represent increased use of the vitamin due to elevated levels of oxidative stress or decreased level of GSH, as GSH is required for recycling vitamin C.

Lipid peroxidation was higher in female N rats when compared with male N rats, whereas a reverse trend was observed in protein carbonyl content and GSH content. The rats with STZ-induced diabetes of both sexes showed

significantly enhanced LPO, Protein oxidation (POD), and decreased GSH content. Administration of AV gel restored the LPO and POD levels to normal in the DT rats of both genders, whereas the GSH was restored to normal levels in male rats and beyond the normal levels in female rats, indicating the protective efficacy of AV gel against oxidative stress.

SOD protects the cells from oxygen-free radicals by catalyzing the removal of superoxide radical, which damages the membrane and biological structures. CAT was shown to be responsible for the detoxification of  $H_2O_2$ .<sup>26</sup> In this study, gender variation was observed in the activities of SOD and CAT, which were significantly higher in male N rats when compared with female N rats. Further, we observed decreased SOD and CAT activities in diabetic rats. This may result in many deleterious effects due to accumulation of superoxide radical and  $H_2O_2$ . The observed increase in antioxidant activities and decline in the activities of TBARSs and hydroperoxides in AV gel extract-treated diabetic rats suggest its potential antilipid peroxidative and antioxidant effects.

Table 4  
Effect of *Aloe vera* treatment on lens polyol pathway enzymes in female rats with STZ-induced diabetes cataract.

Parameter	Tissue	Female rats			
		N	NT	DU	DT
Aldose reductase ( $\mu$ mol of NADPH oxidized/min/mg protein)	Cataractous lens	1.66 ± 0.20	1.70 ± 0.22	6.49 ± 0.21*	1.95 ± 0.12**
Sorbitol dehydrogenase ( $\mu$ mol of NADH oxidized/min/mg protein)	Cataractous lens	1.61 ± 0.09	1.59 ± 0.22	1.02 ± 0.26*	1.59 ± 0.10**

Values are presented as mean ± standard error of the mean for 10 rats in each group.

\* Significant as compared with Group DU ( $p < 0.05$  DMRT).

\*\* Significant as compared with Group DT ( $p < 0.05$  DMRT).

NADH = nicotinamide adenine dinucleotide-reduced; NADPH = nicotinamide adenine dinucleotide phosphate-reduced; STZ = streptozotocin.

Table 5  
Effect of *Aloe vera* treatment on lens protein content in male rats with STZ-induced diabetes cataract.

Parameter	Tissue	Male rats			
		N	NT	DU	DT
Soluble protein (mg/g tissue)	Cataractous lens	118.5 ± 6.77	132.0 ± 5.62	82.8 ± 9.24	186.8 ± 6.12*
Insoluble protein (mg/g tissue)	Cataractous lens	265.2 ± 20.23	256.5 ± 25.70	190.6 ± 26.2**	263.5 ± 13.10*

STZ = streptozotocin.

Values are presented as mean ± standard error of the mean for 10 rats in each group.

\* Significant as compared with Group DT ( $p < 0.05$  DMRT).

\*\* Significant as compared with Group DU ( $p < 0.05$  DMRT).

Table 6  
Effect of *Aloe vera* treatment on lens protein content in female rats with STZ-induced diabetes cataract.

Parameter	Tissue	Female rats			
		N	NT	DU	DT
Soluble protein (mg/g tissue)	Cataractous lens	174.9 ± 4.82	234.1 ± 5.84	45.5 ± 3.93*	156.7 ± 4.72**
Insoluble protein (mg/g tissue)	Cataractous lens	246.36 ± 21.23	236.90 ± 9.84	222.12 ± 14.81	287.00 ± 6.48**

STZ = streptozotocin.

Values are presented as mean ± standard error of the mean for 10 rats in each group.

\*Significant as compared with Group DU ( $p < 0.05$  DMRT).

\*\*Significant as compared with Group DT ( $p < 0.05$  DMRT).

The early accumulation of polyol in the lens leading to the formation of cataract indicates that the polyol pathway plays an important role in the pathogenesis of diabetic cataract. Furthermore, these results are in agreement with previous results in other sugar cataract animal models.<sup>27,28</sup> Sex variation was observed in the activities of polyol pathway enzymes. In this study, the activity of AR increased and that of SDH decreased in Group DU animals compared with Group N in both genders, indicating oxidative stress. Significantly higher AR and SDH activities were observed in male N rats when compared with female N rats. Therefore, prevention of sorbitol accumulation by inhibiting the activity of AR is one possible way to prevent hyperglycemia-induced cataract.<sup>29</sup>

The decrease in the content of soluble and insoluble proteins in Group DU animals compared with Group N lenses in this study could be due to partial leakage of proteins and insolubilization. Treatment with AV gel prevented the decrease in soluble and insoluble proteins in both genders. Nevertheless, the results of this study show that in many respects, the STZ-induced hyperglycemic response of female rats differs from that of male rats described in earlier studies. It is possible that hormonal differences between male and female rats may be one of the factors responsible for the distinct gender difference. Epidemiological evidence suggests that estrogens may protect against cataracts. Although women are at a higher risk of developing cataracts than men, this increased risk comes after menopause, that is, when estrogen levels have waned.<sup>30,31</sup> In a study of 544 women, early onset of menopause was associated with a 2.9-fold risk of developing cataracts.<sup>32</sup> Moreover the results of three small epidemiological studies suggest that postmenopausal estrogen replacement therapy reduces the incidence of cataracts.<sup>33–35</sup>

In conclusion, the results of this study showed that the AV gel extract exhibits antioxidant properties, which may contribute toward preventing peroxidative damage. The gel provides a

viable food-based and a pharmacological approach to the treatment of complications in diabetes. Therefore, delaying cataract formation by treatment with AV gel merits further attention.

## Conflicts of interest

The authors have no conflicts of interest to report.

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